autophagy was attenuated, in peripheral blood mononuclear cells from a patient with a mutation in PIK3CD/p110δ, which is associated with overactivation of mTOR (Lucas et al., 2014). Finally, treatment with rapamycin stimulated autophagy and decreased IL1B secretion in the patient's cells. These results suggest that autophagy regulation by the axis comprised of TORC1 and the mRNA decapping complex is involved in fungal infection, the immune response, and the pathogenesis of human diseases.

Thus, Hu et al. revealed a highly conserved post-transcriptional mechanism in which autophagy gene transcripts are destabilized by a decapping enzyme complex activated by TORC1 under nutrient-rich conditions. Starvation terminates this degradation pathway through inactivation of TORC1. In concert with previously described mechanisms that upregulate transcription of *ATG* genes in response to starvation, this mechanism should allow cells to rapidly induce autophagy under these conditions. Although Dcp2 is the direct target of TORC1-mediated phosphorylation, it is Dhh1/Vad1/DDX6 that associates with substrate mRNAs. Thus, further analysis will be required to elucidate how TORC1-dependent phosphorylation leads to the association of the Dcp2-Dhh1/Vad1/DDX6 complex with *ATG* transcripts. Features in *ATG* transcripts that are recognized by this complex also remain to be determined. Although this study focused on *ATG* genes, whose products drive autophagosome formation, other gene transcripts encoding proteins involved in other steps in autophagy, including autophagosome-lysosome/vacuole fusion and degradation in lysosomes/vacuoles, may also be targets for this degradation pathway. One or more phosphatases that dephosphorylates Dcp2 to accelerate inactivation of this pathway may also exist. Future studies should address these interesting issues raised by this study.

ACKNOWLEDGMENTS

This work was supported by Grants-in-Aid for Scientific Research 25111003, 25711005, and 25111001 from the Ministry of Education, Culture, Sports, Science and Technology of Japan.

REFERENCES

Feng, Y., Yao, Z., and Klionsky, D.J. (2015). Trends Cell Biol. *25*, 354–363.

Freeberg, M.A., Han, T., Moresco, J.J., Kong, A., Yang, Y.C., Lu, Z.J., Yates, J.R., and Kim, J.K. (2013). Genome Biol. *14*, R13.

Hu, G., McQuiston, T., Bernard, A., Park, Y.D., Qiu, J., Vural, A., Zhang, N., Waterman, S.R., Blewett, N.H., Myers, T.G., et al. (2015). Nat. Cell Biol. *17*, 930–942.

Lucas, C.L., Kuehn, H.S., Zhao, F., Niemela, J.E., Deenick, E.K., Palendira, U., Avery, D.T., Moens, L., Cannons, J.L., Biancalana, M., et al. (2014). Nat. Immunol. *15*, 88–97.

Mizushima, N., and Komatsu, M. (2011). Cell *147*, 728–741.

Nakatogawa, H., Suzuki, K., Kamada, Y., and Ohsumi, Y. (2009). Nat. Rev. Mol. Cell Biol. *10*, 458–467.

Presnyak, V., and Coller, J. (2013). Biochim. Biophys. Acta *1829*, 817–823.

Schneider, J.L., and Cuervo, A.M. (2014). Curr. Opin. Genet. Dev. *26*, 16–23.

Shi, C.S., Shenderov, K., Huang, N.N., Kabat, J., Abu-Asab, M., Fitzgerald, K.A., Sher, A., and Kehrl, J.H. (2012). Nat. Immunol. *13*, 255–263.

Yang, Z., and Klionsky, D.J. (2010). Nat. Cell Biol. *12*, 814–822.

Neutrophils, Wounds, and Cancer Progression

Emily E. Rosowski[1] and Anna Huttenlocher[1,*]
[1]Departments of Pediatrics and Medical Microbiology and Immunology, University of Wisconsin-Madison, Madison, WI 53706, USA
*Correspondence: huttenlocher@wisc.edu
http://dx.doi.org/10.1016/j.devcel.2015.07.005

Chronic inflammation is associated with tumorigenesis, but how acute inflammation affects the tumor microenvironment is less known. Recently, Antonio et al. (2015) found that neutrophils attracted to an acute wound such as a biopsy drive cell proliferation of nearby pre-neoplastic cells, suggesting that acute wounds may promote cancer progression.

Cancer is often described as an "unhealed wound," as chronic inflammation is a hallmark of the tumor microenvironment. In fact, unresolved inflammation can promote tumorigenesis—there are well-documented links, for example, between chronic *H. pylori* infection and stomach cancer, inflammatory bowel disease and colon cancer, and chronic hepatitis infection and hepatocellular carcinoma. In other cases, however, immunity and inflammation have beneficial effects and can control tumor growth. Neutrophils are first responders to sites of acute tissue damage, but they are also present in chronic wounds and in the tumor microenvironment, and they have both pro- and anti-tumor functions (Tecchio et al., 2013). There remain gaps in our understanding of how inflammation influences the tumor microenvironment and what factors promote the beneficial or detrimental effects of neutrophils. In particular, how do acute wounds, such as tumor biopsies, which are commonly performed during the diagnosis of cancer, affect cancer progression? This question, and the role of neutrophils in this process, is elegantly addressed in work published recently by Antonio et al. (2015) that used a zebrafish model of RasG12V-induced neoplasia in skin cells (Feng et al., 2010, 2012; Michailidou et al., 2009).

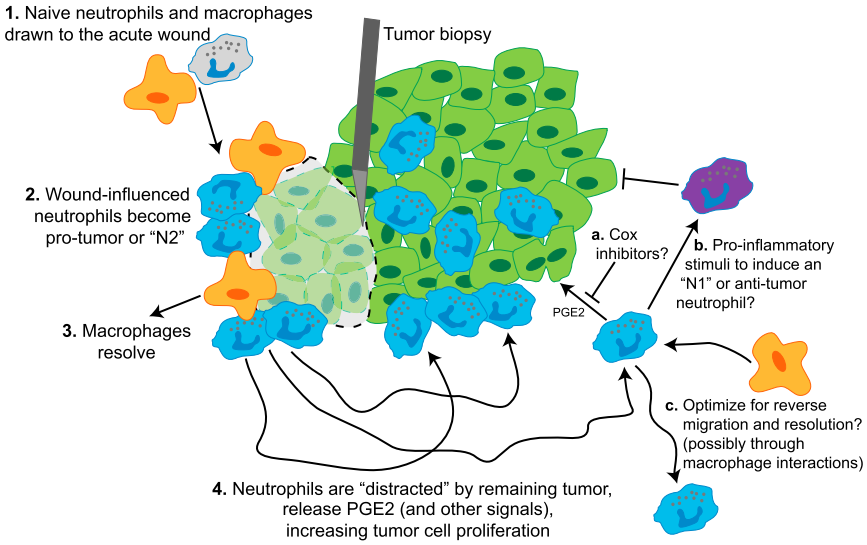

Figure 1. Schematic of Neutrophil-Transformed Cell Interactions after Acute Wounding
After an acute injury, both neutrophils and macrophages infiltrate the wound (1). Unknown signals at the wound influence neutrophils to adopt an anti-inflammatory "N2" phenotype (2). As the wound heals, macrophage inflammation resolves (3), but Antonio et al. find that, if there are nearby cancer cells (as in a tumor biopsy), the N2 neutrophils are "distracted" by and interact with these nearby cancer cells (4). The presence of these neutrophils promotes proliferation of the cancer cells through signals such as PGE2, creating a pro-tumor microenvironment. This suggests that biopsies could be detrimental to the treatment of cancer patients. Future research should focus on how the phenotype of these neutrophils could be optimized to instead inhibit cancer progression. Pro-growth and trophic signals could be directly inhibited by treatment with Cox inhibitors such as aspirin (a), neutrophils could be pushed toward an "N1" or anti-tumor phenotype by treatment with pro-inflammatory stimuli such as LPS (b), or treatments could promote the reverse-migration and resolution of these neutrophils, either directly or by activating subsets of macrophages that repel neutrophils from the wound (c).

Expression of oncogenic RasG12V in zebrafish skin melanocytes or epidermal cells induces cell transformation and recruitment of neutrophils, which drive cell proliferation (Feng et al., 2010) and expression of EMT markers (Freisinger and Huttenlocher, 2014). In the current study, Antonio et al. first demonstrate that chronic tissue damage drives cancer progression; repeatedly wounding adult zebrafish expressing RasG12V in melanocytes significantly increased tumor formation. Turning their attention to the effect of an acute wound in an already chronically "wounded" tumor microenvironment, Antonio et al. found that biopsying sections of these skin tumors increased neutrophil infiltration around neighboring RasG12V$^+$ cells.

To more carefully quantify the link between acute wound-induced inflammation and pre-neoplastic cell growth, Antonio et al. utilized larval zebrafish, where the tumor microenvironment can be imaged in real time. The authors observed that neutrophils migrating to a nearby laser wound were "distracted" by RasG12V$^+$ cells, such that twice as many RasG12V$^+$ cells adjacent to a wound are visited by neutrophils compared to RasG12V$^+$ cells in an unwounded area, and these neutrophils develop sustained interactions with the RasG12V$^+$ cells. Importantly, this acute wound increased the proliferation of nearby existing clones of RasG12V$^+$ cells. Unlike in an unwounded setting, in which the chronic inflammation of both macrophages and neutrophils promotes the growth of these pre-neoplastic cells (Feng et al., 2012), this acute wound-induced proliferation was specifically dependent on neutrophil infiltration: neutrophil depletion with a morpholino against GCSF, but not macrophage depletion with an irf8 morpholino, inhibited this proliferation. It should be noted, however, that depletion of GCSF might independently affect tumor growth, since some cancer cells upregulate the GCSF receptor (Liongue et al., 2009). Thus, these results should be confirmed in other neutrophil-depleted settings.

How do wound-sensitized neutrophils promote tumor progression? What signals are they delivering to these RasG12V cells? Based on previous work, the authors focused on prostaglandin-E2 (PGE2) as a possible trophic signal (Feng et al., 2012). Application of a PGE2 analog in the context of immune depletion partially rescued RasG12V$^+$ cell proliferation. Additionally, treatment with a Cox-2 inhibitor, which dampens the production of PGE2, partially inhibited wounding-induced cell proliferation, suggesting that Cox inhibitors, including non-steroidal anti-inflammatories (NSAIDs) and aspirin, could limit the pro-tumor effect of an acute wound in patients post-biopsy. PGE2 may also be involved in signaling to neutrophils because PGE2 affects the function and behavior of many immune cells.

Larval zebrafish lack an adaptive immune response, allowing Antonio et al. to focus specifically on the role of innate immune cells in driving cancer growth near an acute wound. However, both neutrophils and macrophages can activate (or suppress) T cells and other adaptive immune cells, and it will be interesting in future studies to determine whether the role of neutrophils and macrophages is altered in adult zebrafish or other organisms with an adaptive immune response. In this study the authors found that, as in larval zebrafish, the level of neutrophil, but not macrophage, infiltration in human patient melanoma samples was correlated with tumor cell proliferation. In fact, neutrophil infiltration can be used as a prognostic indicator for patients and is strongly correlated with ulceration status of the melanoma, another known prognostic indicator.

Overall, these findings suggest that acute wounds, such as biopsies, recruit more neutrophils to a tumor site, which in turn promotes proliferation of pre-neoplastic cells (Figure 1). What is different about a neutrophil after visiting an acute wound that makes it even more detrimental in a chronic tumor environment? Future research should focus on how to alter the wound- and tumor-associated neutrophil phenotype to fight, instead of promote, tumor growth. Encouragingly, these data suggest that the effect of an acute wound on pre-neoplastic cell proliferation can be mitigated by treatment with Cox inhibitors such as aspirin. More directed treatments to promote resolution of neutrophil inflammation should also be explored, including inducing neutrophils to leave the wound and reverse-migrate back

into the vasculature (Mathias et al., 2006; Robertson et al., 2014). Targeting macrophages to induce neutrophil resolution or clearance is an intriguing possibility, as subsets of macrophages have been shown to repel neutrophils from a wound (Tauzin et al., 2014). Another possibility is to harness neutrophil inflammation by driving neutrophils toward an anti-tumor "N1" phenotype instead of a pro-tumor "N2" one. How does a nearby infection alter the phenotype of neutrophils in response to a tumor? Application of LPS or other bacterial products could be used to alter the tumor microenvironment to a more anti-cancer function. These questions are very relevant to the clinical setting, where increased tumor growth might be an unintended consequence of a biopsy.

REFERENCES

Antonio, N., Bønnelykke-Behrndtz, M.L., Ward, L.C., Collin, J., Christensen, I.J., Steiniche, T., Schmidt, H., Feng, Y., and Martin, P. (2015). EMBO J., in press. Published online July 1, 2015.

Feng, Y., Santoriello, C., Mione, M., Hurlstone, A., and Martin, P. (2010). PLoS Biol. *8*, e1000562.

Feng, Y., Renshaw, S., and Martin, P. (2012). Curr. Biol. *22*, 1253–1259.

Freisinger, C.M., and Huttenlocher, A. (2014). PLoS ONE *9*, e112183.

Liongue, C., Wright, C., Russell, A.P., and Ward, A.C. (2009). Int. J. Biochem. Cell Biol. *41*, 2372–2375.

Mathias, J.R., Perrin, B.J., Liu, T.X., Kanki, J., Look, A.T., and Huttenlocher, A. (2006). J. Leukoc. Biol. *80*, 1281–1288.

Michailidou, C., Jones, M., Walker, P., Kamarashev, J., Kelly, A., and Hurlstone, A.F.L. (2009). Dis. Model. Mech. *2*, 399–411.

Robertson, A.L., Holmes, G.R., Bojarczuk, A.N., Burgon, J., Loynes, C.A., Chimen, M., Sawtell, A.K., Hamza, B., Willson, J., Walmsley, S.R., et al. (2014). Sci. Transl. Med. *6*, 25ra29.

Tauzin, S., Starnes, T.W., Becker, F.B., Lam, P.-Y.Y., and Huttenlocher, A. (2014). J. Cell Biol. *207*, 589–598.

Tecchio, C., Scapini, P., Pizzolo, G., and Cassatella, M.A. (2013). Semin. Cancer Biol. *23*, 159–170.